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10/576,684	02/06/2007	Thomas Greiner-Stoffele	100728-56-WCG	4662
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BOESEN, CHRISTIAN C				
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary

Application No.

10/576,684

Applicant(s)

GREINER-STOFFELE ET AL.

Examiner

CHRISTIAN BOESEN

Art Unit

1639

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --
Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

- 1) ☒ Responsive to communication(s) filed on 27 August 2009.
- 2a) ☒ This action is **FINAL**. 2b) ☐ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

- 4) ☒ Claim(s) 1-15 is/are pending in the application.
- 4a) Of the above claim(s) _____ is/are withdrawn from consideration.
- 5) ☐ Claim(s) _____ is/are allowed.
- 6) ☒ Claim(s) 1-15 is/are rejected.
- 7) ☐ Claim(s) _____ is/are objected to.
- 8) ☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☐ The drawing(s) filed on _____ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some * c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
 2. ☐ Certified copies of the priority documents have been received in Application No. _____.
 3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

- 1) ☒ Notice of References Cited (PTO-892)
- 2) ☐ Notice of Draftperson's Patent Drawing Review (PTO-948)
- 3) ☐ Information Disclosure Statement(s) (PTO/SB/CD)
Paper No(s)/Mail Date _____
- 4) ☐ Interview Summary (PTO-413)
Paper No(s)/Mail Date _____
- 5) ☐ Notice of Informal Patent Application
- 6) ☐ Other: _____

DETAILED ACTION

This Final Office Action is responsive to the communication received 08/27/2009.

Claim Status

Claim(s) 8, 11 and 15 have been amended as filed on 08/27/2009.

Claim(s) 1-15 are currently pending.

Claim(s) 1-15 are being examined in this application.

Election/Restrictions

Applicant's election in the application filed on 04/20/2006 of group I (election by original presentation, no restriction filed), claims 1-15 is noted.

Priority

This application for patent is filed under 35 U.S.C 371 of PCT/DE04/02386 (filed on 10/22/2004).

Acknowledgment is made of applicant's claim for foreign priority based on an application filed in Germany on 10/23/2003. The applicant has filed a certified copy of the 103 50 474.5 application as required by 35 U.S.C. 119(b).

Claim Rejection(s) Withdrawn

Upon further consideration and in light of Applicant's arguments and/or amendments, the following claim rejection(s) as set forth in the previous office action is(are) withdrawn:

1. Claims 8, 10-11, and 15 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim Rejection(s) Maintained

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless –

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 1-5 and 7 and 9 and 12 and 14 are rejected under 35 U.S.C. 102(b) as being anticipated by Hubner (Biochemistry, 1999, volume 38, page 1371).

Claims are drawn to a method for the identification of biomolecules in variant libraries.

Hubner discloses **claim 1(a)**, the production of variant library (page 1373 left column, center, "For library construction, we randomized the polypeptide segment between positions 42 and 46, which is mainly involved in guanine binding.")

Hubner discloses **claim 1(b)**, the division of the variant library by streaking the library out on agar plates with indicators (page 1372, right column, center, "Competent *E. coli*

DH5 α cells were prepared by the method of Inoue et al. (31), yielding a transformation efficiency of 5×10^7 colony forming units/ μ g of pBR322. Cells were transformed with 40 ng of the ligation mixture per 100 μ L cell aliquot. Transformants were screened for RNA hydrolysis activity on RNase indicator plates (32) containing Luria-Bertani medium supplemented with 100 mg of ampicillin, 75 mg of toluidine blue O, and 2 g of yeast RNA per liter.")

Hubner discloses **claim 1(c)**, the production of biomolecules in the transformed bacteria (page 1373, right column, center, "RNase variants were expressed and secreted by *E. coli* cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32).")

Hubner discloses **claim 1(d)**, the selection of biomolecules on agar plates (page 1372, right column, center, "Colonies secreting active RNase variants could be identified by red haloes.")

Hubner discloses **claim 1(e) and (f)**, that the library can be divided and subjected to additional rounds of screening (page 1373, right column, center, "In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants.")

Hubner discloses **claim 2**, selection for biocatalytic activity (page 1373, right column, center, "*Selection of Active Enzymes and Determination of Their Base Specificity.*")

Hubner discloses **claim 3**, amplification of the partial library, (page 1373, right column, center, "In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants.")

Hubner discloses **claim 4**, dilution of the partial library (page 1373, right column, center, "In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants.")

Hubner discloses **claim 5**, a variant library contains 10^3 to 10^{15} variants (page 1373, right column, center, "In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants.")

Hubner discloses **claim 7**, the variant library is transferred into an organism before division (page 1373, right column, center, "RNase variants were expressed and secreted by *E. coli* cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32).")

Hubner discloses **claim 9**, the organisms also conduct the production of the biomolecules (page 1373, right column, center, "RNase variants were expressed and secreted by *E. coli* cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32).")

Hubner discloses **claim 12**, a variant library consists of DNA-plasmids, which contain the gene sequence coding for the biomolecule (page 1373, right column, center, "RNase variants were expressed and secreted by *E. coli* cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32).")

Hubner discloses **claim 14**, the biomolecules are enzymes or ribozymes or other biomolecules, which exhibit a biocatalytic activity (page 1373, right column, bottom, "The hydrolysis profiles analyzed indicated that the RNase T1 variant RNase T1-8/3 (K41S/N43W/N44H/Y45A/E46D) was cleaving substrates with guanine and adenine in the 5' position, whereas all other variants still exhibited exclusively guanine specificity.")

Thus, by this disclosure Hubner anticipates the present claims.

Discussion and Answer to Argument

Applicant's arguments have been fully considered but they are not persuasive for the following reasons (in addition to reasons of record). Each point of Applicant's traversal is addressed below (Applicant's arguments are in italic):

1. Applicant asserts "*In contrast, in Applicants' method step b), the variants are divided into compartments that, unlike Hubner et al., intentionally comprise multiple variants, B₀/W₀.*" (Reply page 7 top).

In response to Applicant's arguments, the Examiner respectfully disagrees.

With regards to **claim 1(a)**, Hubner discloses production of a variant library, consisting of a number of variants (B_0) of gene sequences coding for the biomolecule, for example

"For **library** construction, we randomized the polypeptide segment between positions 42 and 46, which is mainly involved in guanine binding." (page 1373 left center) and "Site-directed random mutagenesis was carried out by a two-step PCR technique (27). Primers used were A2vo (5'-TAC GGA TTC ACT GGA ACT C-3'), A2hi (5'-CAT CTT AGC AGC CTG AAC-3'), and the mutagenesis primer 41-46* (5'-CAC AGA GAA ATC AAA ACC SNN SNN SNN SNN SNN SNN GTG TGG GTA AGA ATT GG-3'), where N means an equimolar mixture of all four nucleotides and S a mixture of 50% G and C each. In the first PCR step, 100 ng of DNA from plasmid pA2T1 (28, 29) or its derivative coding for the inactive variant, the universal primer A2vo, and the random mutagenesis primer 41-46* were applied for amplification." (page 1372 left bottom) and "Transformation of competent *E. coli* DH5 α F" with an aliquot of the ligation mixture resulted in a total of 3.2×10^6 independent transformants with a vector background from self-ligation of less than 1%." (page 1373 right top) and "In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

The library is the randomized the polypeptide segment between positions 42 and 46. The library contains 5 residues that are randomized by NNS mutagenesis. The NNN codon complexity is 64. The NNS codon complexity is 32. If the library is grown in cells with an amber mutation the stop codon is not functional the thus, the 32 codons code for a total of 20 different amino acid residues. Theoretically, $B_0 = 20^5 = 3.2 \times 10^6$ gene sequences coding for the biomolecule. The actual $B_0 = 3.2 \times 10^6$ or $B_0 = 1.6 \times 10^6$ for two different libraries.

With regards to **claim 1(b)**, the recitation "compartment" is not defined in the specification and is therefore given the broadest reasonable definition of "a separate division or section" from Merriam Websters Online Dictionary (downloaded 12/22/2009 from <http://www.merriam-webster.com/dictionary>) main entry for compartment.

With regards to **claim 1(b)**, Hubner discloses division of the variant library into a number of compartments (W_0), which is at least by a factor of ten smaller than the number of variants in the variant library (B_0), where each compartment contains a partial library which contains $K_0=B_0/W_0$ variants, for example

"RNase variants were expressed and secreted by E. coli cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32). Thus, large numbers of variants could be tested for RNA hydrolysis activity by colony screening. In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

In the case transformants tested for the RNase indicator, Hubner tests transformants on plates. The area of the lawn of transformants that contain a positive red halo that can be picked is a compartment = (W_0). Bacteria from picked area is grown in a liquid media with ampicillin, then DNA plasmid isolated, then transformed and plated again for selection. The lawn is not an individual colony on the first plating and thus requires several rounds of transformation and plating to identify one individual unique clone.

2. *Applicant asserts "Those compartments are then assayed in step c) for a particular phenotype. If a phenotype is detected in one of those compartments, one of skill in the art cannot assign that detected phenotype to a particular genotype because that compartment comprises multiple variants: the genotype and phenotype in step c) are therefore "uncoupled." Hubner et al. do not disclose the method according to claim 1, wherein genotype and phenotype are "uncoupled."" (Reply page 7 top).*

In response to Applicant's arguments, the Examiner respectfully disagrees.

With regards to **claim 1(c)**, Hubner discloses production of biomolecules in the compartments and testing of the biomolecules obtained in the single compartments for a specified phenotype, whereas from the observed phenotype no direct conclusions on the genotype can be made, for example

"RNase variants were expressed and secreted by E. coli cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32). Thus, large numbers of variants could be tested for RNA hydrolysis activity by colony screening. In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

In the case transformants tested for the RNase indicator, Hubner tests transformants on plates. The area of the lawn of transformants that contain a positive red halo that can be picked is a compartment = (W_0). Bacteria from picked area is grown in a liquid media with ampicillin, then DNA plasmid isolated, then transformed and plated again for selection. The lawn is not an individual colony on the first plating and thus requires several rounds of transformation and plating to identify one individual unique clone. In the first round of plating the positive phenotype (red halo) does not correspond to the positive phenotype because the compartment has a mixture of positive and negative genotypes and phenotypes. Thus, additional rounds of transformation and plating are required to identify a single clone. The single clone is the one positive colony with a coupled genotype and phenotype. Before the individual clone is isolated the genotype and phenotype are uncoupled.

3. *Applicant asserts "In step d) of the claimed method the compartment having the detected phenotype is then selected and in step e) the variants further divided into more compartments each compartment having a number of variants. Then the new compartments are assayed for the desired phenotype, step c). Steps c) through e) are repeated until the compartments contain, at a maximum, one variant. Once the compartments contain maximally one variant, and the desired phenotype is detected in a compartment, the detected phenotype can be assigned to the genotype of the variant in that compartment." (Reply page 7 center).*

In response to Applicant's arguments, the Examiner respectfully disagrees.

With regards to **claim 1(d)**, Hubner discloses selection of at least one compartment, which contains biomolecules fulfilling the wanted properties, for example

"RNase variants were expressed and secreted by E. coli cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32). Thus, large numbers of variants could be tested for RNA hydrolysis activity by colony screening. In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

Selection is the picking of positive area with red halos before each round of transformation and plating.

With regards to **claim 1(e)**, Hubner discloses division of the partial library contained in the selected compartment into further compartments, for example

"RNase variants were expressed and secreted by E. coli cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32). Thus, large numbers of variants could be tested for RNA hydrolysis activity by colony screening. In several rounds of transformation and plating on RNase indicator plates, 180

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active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

Each round of selection, transformation and plating involves a new division of the partial library.

With regards to **claim 1(f)**, Hubner discloses n-fold repetition of the steps c) to c) until in every compartment maximally only one variant ($K_n \leq 1$) of the gene sequence coding for the biomolecule is contained, for example

"RNase variants were expressed and secreted by E. coli cells using the expression/secretion vector pA2T1, and active RNases could be detected on RNase indicator plates (32). Thus, large numbers of variants could be tested for RNA hydrolysis activity by colony screening. In several rounds of transformation and plating on RNase indicator plates, 180 active variants were detected from a total number of 1.6×10^6 independent transformants." (page 1373 right center).

After multiple rounds 180 individual clones were identified. Each clone contained a single gene coding for the biomolecule (RNase).

Summary

Hubner teaches a library of active and inactive RNase T1 enzymes, thus the phenotype (active RNase) and the genotype (DNA coding for active RNase) and uncoupled in the library. Hubner teaches using multiple rounds of transformation of the library of plasmids containing DNA coding for active or inactive RNase, screening by plating and identifying red halos, selection by picking bacteria from the region of the plate with red halos and growing the selected bacteria in a culture followed by isolation of the plasmid containing the DNA coding for active or inactive RNase. Rounds of this transformation, plating, and selection are required to identify

individual clones and couple the phenotype (formation of red halos) with the genotype (DNA coding for an active RNase).

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

Claims 6, 8, 11, and 13 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hubner (Biochemistry, 1999, volume 38, page 1371) as applied to claims 1 and 7 in combination with Selfonov (WO 01/12791 A1).

Claims are drawn to a method for the identification of biomolecules in variant libraries.

While Hubner teaches a method for the identification of biomolecules in variant libraries, as discussed above, Hubner does not teach that the variant library is divided up in 10^1 to 10^4

compartments, or that the culture of the organism after division is amplified to a number of organisms of 10^8 to 10^9 per compartment, or the partial libraries in the compartments are isolated from the organisms, or the amplification of the partial libraries and the production of the biomolecules is conducted by cell-free systems, or the variant library consists of linear nucleic acid molecules, which contain the gene sequence coding for the biomolecule.

Stelifonov teaches **claim 6**, the variant library is divided up in 10^1 to 10^4 compartments (page 95, line 34, "To increase the chances of identifying a pool of sufficient size, a prescreen that increases the number of mutants processed by 10-fold can be used. The goal of the primary screen will be to quickly identify mutants having equal or better product titers than the parent strain(s) and to move only these mutants forward to liquid cell culture for subsequent analysis.")

Stelifonov teaches **claim 8**, that the culture of the organism after division is amplified to a number of organisms of 10^8 to 10^9 per compartment (page 40, line 3, "The starting DNA segments are recombined by any of the sequence recombination formats provided herein to generate a diverse library of recombinant DNA segments. Such a library can vary widely in size from having fewer than 10 to more than 10^5 , 10^9 , 10^{12} or more members.")

Stelifonov teaches **claim 11**, the amplification of the partial libraries is conducted by cell-free systems (page 22, line 20, "Alternatively, initial diversity can be induced, e.g., the variant forms can be generated by error-prone transcription, such as an error-prone PCR or use of a polymerase which lacks proof-reading activity (see, Liao (1990) Gene 88: 107-111), of the first

variant form, or, by replication of the first form in a mutator strain (mutator host cells are discussed in further detail below, and are generally well known)." and page 5 line 24, "If sequence recombination is performed *in vitro*, the recombinant library is preferably introduced into the desired cell type before screening/selection.")

Stelifonov teaches **claim 13**, the variant library consists of linear nucleic acid molecules (the products of PCR), which contain the gene sequence coding for the biomolecule (page 22, line 20, " Alternatively, initial diversity can be induced, e.g., the variant forms can be generated by error-prone transcription, such as an error-prone PCR or use of a polymerase which lacks proof-reading activity (see, Liao (1990) Gene 88: 107-111), of the first variant form, or, by replication of the first form in a mutator strain (mutator host cells are discussed in further detail below, and are generally well known).")

The present claims would have been obvious because the **substitution** of one known element 10-fold, taught by Stelifonov for another (quantity not specified), taught by Hubner would have yielded predictable results to one of ordinary skill in the art at the time of the invention (i.e. a 10-fold division of the variant library). See *KSR International Co. v. Teleflex Inc.*, 82 USPQ2d 1385 (U.S. 2007).

The present claims would have been obvious because the **substitution** of one known element 10 to more than 10^5 , 10^9 , 10^{12} or more members, taught by Stelifonov for another 1.6×10^6 , taught by Hubner would have yielded predictable results to one of ordinary skill in the art at

the time of the invention (i.e. a library of organisms of 10^8 to 10^9). See *KSR International Co. v. Teleflex Inc.*, 82 USPQ2d 1385 (U.S. 2007).

The present claims would have been obvious because the **substitution** of one known element *in vitro*, taught by Stelifonov for another *in vivo*, taught by Hubner would have yielded predictable results to one of ordinary skill in the art at the time of the invention (i.e. *in vitro* amplification of libraries). See *KSR International Co. v. Teleflex Inc.*, 82 USPQ2d 1385 (U.S. 2007).

The present claims would have been obvious because the **substitution** of one known element linear nucleic acid, taught by Stelifonov for another vector nucleic acid, taught by Hubner would have yielded predictable results to one of ordinary skill in the art at the time of the invention (i.e. linear nucleic acid libraries). See *KSR International Co. v. Teleflex Inc.*, 82 USPQ2d 1385 (U.S. 2007).

All the claimed elements were known in the prior art and one skilled in the art could have combined the elements as claimed by known methods with no change in their respective functions, and the combination would have yielded predictable results to one of ordinary skill in the art at the time of the invention.

Thus, the present invention would have been *prima facie* obvious at the time the invention was made.

Discussion and Answer to Argument

Applicant's arguments have been fully considered but they are not persuasive for the following reasons (in addition to reasons of record). Each point of Applicant's traversal is addressed below (Applicant's arguments are in *italic*):

Applicants traversed the rejections over the Hubner (Biochemistry, 1999, volume 38, page 1371) reference with the same argument as the traversal over the combination of the Hubner (Biochemistry, 1999, volume 38, page 1371) in combination with Selifonov (WO 01/12791 A1) references. Thus, applicants are respectfully directed to the above discussion for answer to arguments.

Claim 10 is rejected under 35 U.S.C. 103(a) as being unpatentable over Hubner (Biochemistry, 1999, volume 38, page 1371) as applied to claims 1 and 7 in combination with Napolitano (The Journal of Neuroscience, 1987, volume 7, page 2590).

Claims are drawn to a method for the identification of biomolecules in variant libraries.

While Hubner teaches a method for the *in vivo* expression of biomolecules, as discussed above, Hubner does not teach the *in vitro* expression of biomolecules.

Napolitano teaches **claim 10**, the *in vitro* protein expression of biomolecules starting with a cDNA clone (page 2595, left column, bottom, "NF-M mRNA was transcribed *in vitro* using the pNF-M2D insert subcloned into the pGEM system.")

The present claims would have been obvious because the **substitution** of one known element *in vitro* protein expression, taught by Napolitano for another *in vivo* protein expression, taught by Hubner would have yielded predictable results to one of ordinary skill in the art at the

time of the invention (i.e. *in vitro* expression of biomolecules). See *KSR International Co. v. Teleflex Inc.*, 82 USPQ2d 1385 (U.S. 2007).

All the claimed elements were known in the prior art and one skilled in the art could have combined the elements as claimed by known methods with no change in their respective functions, and the combination would have yielded predictable results to one of ordinary skill in the art at the time of the invention.

Thus, the present invention would have been *prima facie* obvious at the time the invention was made.

Discussion and Answer to Argument

Applicant's arguments have been fully considered but they are not persuasive for the following reasons (in addition to reasons of record). Each point of Applicant's traversal is addressed below (Applicant's arguments are in *italic*):

Applicants traversed the rejections over the Hubner (Biochemistry, 1999, volume 38, page 1371) reference with the same argument as the traversal over the combination of the Hubner (Biochemistry, 1999, volume 38, page 1371) in combination with Napolitano (The Journal of Neuroscience, 1987, volume 7, page 2590) references. Thus, applicants are respectfully directed to the above discussion for answer to arguments.

Claim 15 is rejected under 35 U.S.C. 103(a) as being unpatentable over Hubner (Biochemistry, 1999, volume 38, page 1371) as applied to claim 1 in combination with Korn (Methods in Enzymology, 2001, volume 341, page 142).

Claims are drawn to a method for the identification of biomolecules in variant libraries.

While Hubner teaches a method for the identification of biomolecules in variant libraries, as discussed above, Hubner does not teach that the test for a biocatalytic activity is conducted with fluorescence correlation spectroscopy.

Korn teaches **claim 15**, that the test for a biocatalytic activity is conducted with fluorescence correlation spectroscopy (page 142, top, "To investigate ribonuclease (RNase)-mediated cleavage of its presumed natural substrates - long RNA molecules - a variety of different assays have been developed" and page 142, bottom, "Furthermore, we present two assays to study the degradation of larger RNA substrate molecules using either methylene blue or fluorescence correlation spectroscopy (FCS).")

It would have been *prima facie* obvious for one of ordinary skill in the art at the time the invention was made to use Korn's ribonuclease assay in Hubner's method for the identification of biomolecules in variant libraries **to arrive at applicant's invention with the above cited references before them.**

One would have been motivated to use Korn's ribonuclease assay because Korn teaches "Binding, accumulation, and cleavage processes can be easily followed by FCS bases on the relation of the molecular weight and diffusion times of the molecules." (page 148, middle).

One would have had a reasonable expectation of success to assay ribonuclease activity because this assay is well established in the art as evidenced by Korn (page 151, top, "Table I, Diffusion times of rhodamine B-labeled deoxyribonucleotide (dOligo) and heteroduplexes")

Thus, the present invention would have been *prima facie* obvious at the time the invention was made.

Discussion and Answer to Argument

Applicant's arguments have been fully considered but they are not persuasive for the following reasons (in addition to reasons of record). Each point of Applicant's traversal is addressed below (Applicant's arguments are in italic):

Applicants traversed the rejections over the Hubner (Biochemistry, 1999, volume 38, page 1371) reference with the same argument as the traversal over the combination of the Hubner (Biochemistry, 1999, volume 38, page 1371) in combination with Korn (Methods in Enzymology, 2001, volume 341, page 142) references. Thus, applicants are respectfully directed to the above discussion for answer to arguments.

Conclusion

No claim is allowed.

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR

1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

Any inquiry concerning this communication or earlier communications from the Examiner should be directed to CHRISTIAN BOESEN whose telephone number is 571-270-1321. The Examiner can normally be reached on Monday-Friday 9:00 AM to 5:00 PM.

If attempts to reach the Examiner by telephone are unsuccessful, the Examiner's supervisor, Christopher S. Low can be reached on 571-272-0951. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

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